

IN THE CLAIMS:

This listing of claims replaces all prior versions and listings of claims in the application:

Listing of Claims

1. (Canceled)
2. (Canceled)
3. (Withdrawn) A DNA which is capable of hybridizing to the nucleotide sequence of SEQ ID No: 1 or to the complement thereof under such conditions that the hybridization is carried out in 6xSSC and 50% formamide at 42 ° C and the washing process is carried out in 6xSSC and 40% formamide at 25° C, and which encodes a thermophilic enzyme having β -glycosidase activity.
4. (Withdrawn) The DNA of claim 3, which encodes the enzyme of claim 1.
5. (Withdrawn) A recombinant vector containing the DNA of claim 3 therein.
6. (Withdrawn) A host cell transformed with the recombinant vector of claim 5.
7. (Withdrawn) A process for producing the enzyme of claim 1, comprising culturing a host cell transformed with an expression vector containing a DNA encoding the enzyme and then collecting the enzyme from the resultant culture.

8. (Previously Presented) A process for the hydrolysis of a β -glycoside having a long alkyl chain at the reducing end, comprising contacting the β -glycoside with a thermophilic enzyme comprising the amino acid sequence of SEQ ID NO: 2.

9. (Original) The process of claim 8, wherein the long alkyl chain is an alkyl group having carbon atoms of 8 or more.

10. (Original) The process of claim 8, wherein the hydrolysis is carried out at a temperature of 85 °C or higher.

11. (Original) The process of claim 8, wherein the hydrolysis is carried out at a temperature of 100°C or higher.

12. (Canceled)

13. (Canceled)

14. (Currently Amended) A method for using a thermophilic enzyme as a β -glycosidase, comprising the following steps:

(a) providing an enzyme, wherein the enzyme comprises four subunits to form a tetramer, wherein each subunit of the tetramer comprises a sequence as set forth in SEQ ID NO:2; and

(b) contacting the tetrameric enzyme with a ~~substrate~~ β -glucoside comprising a long alkyl chain under conditions wherein the enzyme functions as a β -glycosidase ~~on the substrate~~.

15. (Canceled)

16. (Currently Amended) The method of claim ~~15~~ 14, wherein the long alkyl chain comprises 8 or more carbon atoms.

17. (Previously presented) The method of claim 14, wherein the enzyme has a high affinity to a β -glucoside comprising a long alkyl chain.

18. (Currently Amended) The method of claim ~~15~~ 14, wherein the β -glucoside having a long alkyl chain is selected from the group consisting of n-Dodecyl- β -D-Glcp and n-Octyl- β -D-Glcp.

19. (Previously presented) The method of claim 14, wherein the function comprises synthesis of an oligosaccharide or a heterosaccharide with optical purity.

20. (Previously presented) The method of claim 14, wherein the conditions comprise temperatures selected from the group consisting of 90°C or higher and 100°C or higher.

21. (Previously presented) The method of claim 14, wherein the conditions comprise an organic solvent.

22. (Previously presented) The method of claim 14, wherein the enzyme is encoded by a nucleotide sequence comprising SEQ ID NO:1.

23. (Previously presented) The method of claim 14, wherein the enzyme is encoded by a nucleotide sequence capable of hybridizing to SEQ ID NO:1, or its complement, under moderately stringent conditions of 6xSSC and 40% formamide at 42°C.

24. (Previously presented) The method of claim 23, wherein the hybridization further comprises a washing step carried out in 1xSSC and 0% formamide at 55°C.

25. (Withdrawn) A method of producing a recombinant vector for the enzyme of claim 14, comprising:

- a) providing a recombinant vector comprising the nucleotide encoding SEQ ID NO:2; and
- b) transforming a host cell with the recombinant vector.

26. (Withdrawn) A method of producing the enzyme of claim 14, comprising:

- a) providing an expression vector comprising the nucleotide encoding SEQ ID NO:2;
- b) transforming a host cell with the expression vector;
- c) culturing the host cell; and
- d) expressing and collecting the enzyme.

27. (Withdrawn) A method of solubilizing the enzyme of claim 14, wherein a solubilization condition comprises heating with about 2.5% Triton X-100 at about 85°C for about 15 min.

28. (Currently Amended) A method for using a β -glycosidase, comprising:
(a) providing a β -glycosidase, wherein the β -glycosidase comprises a tetramer of four subunits, and at least one subunit is encoded by a nucleic acid that hybridizes to a polynucleotide consisting of SEQ ID NO:1, or its complement, under hybridization conditions comprising 6xSSC and 50% formamide at 42°C and washing conditions comprising 6xSSC and 40% formamide at 25°C, and wherein the nucleic acid encodes a subunit of the β -glycosidase and wherein the β -glycosidase is active at temperatures at or above about 90°C or higher; and

(b) contacting the β -glycosidase with a substrate comprising a β -glycoside having a long alkyl chain at the reducing end under conditions wherein the β -glycosidase functions as a β -glycosidase on the substrate.

29. (Canceled)

30. (Currently Amended) A method for using a β -glycosidase, comprising:

(a) providing a β -glycosidase, wherein the β -glycosidase comprises a tetramer of four subunits, and each subunit is encoded by a nucleic acid that hybridizes to a polynucleotide consisting of SEQ ID NO:1, or its complement, under hybridization conditions comprising 6xSSC and 40% formamide at 42°C and washing conditions comprising 1xSSC and 0% formamide at 55°C, and wherein the nucleic acid encodes a subunit of the β -glycosidase and wherein the β -glycosidase is active at temperatures at or above about 90°C or higher; and

(b) contacting the β -glycosidase with a substrate comprising a β -glycoside having a long alkyl chain at the reducing end under conditions wherein the β -glycosidase functions as a β -glycosidase on the substrate.

31. (Currently Amended) A method for using a β -glycosidase, comprising:

(a) providing a β -glycosidase, wherein the β -glycosidase comprises a tetramer of four subunits, and each subunit is encoded by a nucleic acid that hybridizes to a polynucleotide consisting of SEQ ID NO:1, or its complement, under hybridization conditions comprising 6xSSC and 30% formamide at 42°C and washing conditions comprising 0.1xSSC and 0% formamide at 62°C, and wherein the nucleic acid encodes a subunit of the β -glycosidase and wherein the β -glycosidase is active at temperatures at or above about 90°C or higher; and

(b) contacting the β -glycosidase with a substrate comprising a β -glycoside having a long alkyl chain at the reducing end under conditions wherein the β -glycosidase functions as a β -glycosidase on the substrate.

32. (Withdrawn) A method for making a β -glycosidase enzyme, comprising the following steps:

(a) providing four subunits of a tetramer, wherein each subunit is encoded by a nucleic acid comprising a sequence capable of hybridizing to SEQ ID NO:1, or its complement, under conditions comprising a hybridization step comprising 6xSSC and 50% formamide at 42°C and a washing step comprising 6xSSC and 40% formamide at 25°C, and the β -glycosidase is active at temperatures at or above about 90°C or higher; and

(b) contacting the four subunits together under conditions wherein they form a tetrameric enzyme comprising a β -glycosidase activity.

33. (Currently Amended) A method for hydrolyzing a β -glycoside, comprising:

(a) providing a β -glycosidase, wherein the β -glycosidase comprises a tetramer of four subunits, and each subunit is encoded by a nucleic acid that hybridizes to a polynucleotide consisting of SEQ ID NO:1, or its complement, under hybridization conditions comprising 6xSSC and 50% formamide at 42°C and washing conditions comprising 6xSSC and 40% formamide at 25°C, and wherein the nucleic acid encodes a subunit of the β -glycosidase and wherein the β -glycosidase is active at temperatures at or above about 90°C or higher; and

(b) contacting the β -glycosidase with a β -glycoside having a long alkyl chain at the reducing end under conditions wherein the β -glycosidase hydrolyzes the β -glycoside.

34. (Previously Presented) The process of claim 8, wherein the β -glycoside is contacted with the enzyme in 50 mM phosphate buffer (pH 6.0) with 0.1% Triton X-100 and 0.3 M NaCl at 90 °C.